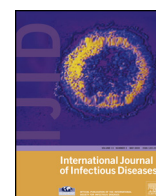


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## Review

Community-acquired pneumonia and tuberculosis: differential diagnosis and the use of fluoroquinolones<sup>☆</sup>Ronald F. Grossman<sup>a,\*</sup>, Po-Ren Hsueh<sup>b</sup>, Stephen H. Gillespie<sup>c</sup>, Francesco Blasi<sup>d</sup><sup>a</sup> University of Toronto, 2300 Eglinton Ave West, Suite 201, Mississauga, Ontario, L5M 2V8, Canada<sup>b</sup> Departments of Laboratory Medicine and Internal Medicine, National Taiwan University Hospital, National Taiwan University College of Medicine, Taipei, Taiwan<sup>c</sup> School of Medicine, University of St Andrews, St Andrews, Fife, UK<sup>d</sup> Department of Pathophysiology and Transplantation, University of Milan, IRCCS Fondazione Cà Granda Milano, Milan, Italy

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## SUMMARY

The respiratory fluoroquinolones moxifloxacin, gemifloxacin, and high-dose levofloxacin are recommended in guidelines for effective empirical antimicrobial therapy of community-acquired pneumonia (CAP). The use of these antibiotics for this indication in areas with a high prevalence of tuberculosis (TB) has been questioned due to the perception that they contribute both to delays in the diagnosis of pulmonary TB and to the emergence of fluoroquinolone-resistant strains of *Mycobacterium tuberculosis*. In this review, we consider some of the important questions regarding the potential use of fluoroquinolones for the treatment of CAP where the burden of TB is high. The evidence suggests that the use of fluoroquinolones as recommended for 5–10 days as empirical treatment for CAP, according to current clinical management guidelines, is appropriate even in TB-endemic regions. It is critical to quickly exclude *M. tuberculosis* as a cause of CAP using the most rapid relevant diagnostic investigations in the management of all patients with CAP.

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## 1. Introduction

The respiratory fluoroquinolones moxifloxacin, gemifloxacin, and levofloxacin (at a daily dose of 750 mg) are recommended for empirical antimicrobial therapy of community-acquired pneumonia (CAP).<sup>1,2</sup> Despite their proven worth in CAP, it has been suggested that fluoroquinolone use should be restricted to the management of tuberculosis (TB), even though there have been few well-controlled clinical studies of their use in TB-endemic parts of the world.<sup>3–6</sup> More specifically, some authors have proposed that newer fluoroquinolones should not be used in areas of TB endemicity, given the potential to mask active TB and the threat of an emerging epidemic of fluoroquinolone- and extensively drug-resistant (XDR) TB.<sup>7,8</sup>

In this review, we examine the role of respiratory fluoroquinolones in the treatment of both TB and CAP and consider how these agents should be used in the context of both infections.

## 2. Fluoroquinolone treatment in the management of CAP

CAP may be caused by a wide variety of pathogens, but a limited number of agents are responsible for most cases. Recent data have confirmed *Streptococcus pneumoniae* to be the most common pathogen isolated from patients with CAP.<sup>1,2</sup> Other bacterial causes include non-typeable *Haemophilus influenzae* and *Moraxella catarrhalis*, generally in patients with underlying bronchopulmonary disease, *Staphylococcus aureus*, especially during an influenza outbreak, and so-called 'atypical' organisms, such as *Mycoplasma pneumoniae*, *Chlamydia pneumoniae*, *Legionella* species, and respiratory viruses.<sup>1,9</sup>

There is good pharmacological and clinical evidence to support the use of respiratory fluoroquinolones in CAP. Their favourable pharmacokinetic and pharmacodynamic profiles result in good penetration of respiratory tissues; the administration of a single 400-mg oral dose of moxifloxacin, for example, achieves higher concentrations in alveolar macrophages (56.7 µg/ml) and

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epithelial lining fluid (20.7 µg/ml) than in serum (3.2 µg/ml).<sup>10</sup> The broad antibacterial activity of respiratory fluoroquinolones provides excellent coverage of the major CAP-causing pathogens, including penicillin- and macrolide-resistant *S. pneumoniae*.<sup>11</sup> Dosing is once daily and the availability of oral and intravenous formulations of moxifloxacin and levofloxacin allows delivery of effective therapy to a wide range of patients, including the critically ill.<sup>2</sup> Ineffective initial therapy of CAP is the most significant prognostic and single intervention-related factor linked to mortality.<sup>12</sup> A meta-analysis of 15 clinical trials showed that pneumonia was cured or improved in significantly more patients treated with fluoroquinolones than those treated with macrolide ± beta-lactam antibiotics.<sup>13</sup> Moxifloxacin monotherapy, for example, has been shown to be superior to amoxicillin-clavulanic acid ± clarithromycin in terms of clinical cure and bacteriological success in the treatment of patients hospitalized with CAP.<sup>14</sup> Fluoroquinolones were also more effective than macrolides ± beta-lactams for patients with severe pneumonia, those who were hospitalized and those who required intravenous therapy.<sup>13</sup>

Fluoroquinolones are generally recommended in different management guidelines for use in CAP, i.e., pneumonia in immunocompetent subjects arising outside of the hospital. The Infectious Diseases Society of America (IDSA) and the American Thoracic Society (ATS) consensus guidelines, for example, recommend monotherapy with a respiratory fluoroquinolone for patients with CAP admitted to general medical wards, or a combination of a beta-lactam and a respiratory fluoroquinolone for patients admitted to intensive care units (ICUs) and who do not have risk factors for methicillin-resistant *S. aureus* or *Pseudomonas spp.*<sup>1</sup> The European Respiratory Society (ERS) and European Society for Clinical Microbiology and Infectious Diseases (ESCMID) guidelines recommend a fluoroquinolone as: (1) first-line monotherapy for hospitalized (non-ICU) patients with CAP; (2) monotherapy or in combination with a non-antipseudomonal cephalosporin for patients with severe CAP in the ICU or intermediate care; and (3) second-choice agent for the treatment of CAP in outpatients.<sup>2</sup> In the treatment of patients hospitalized with CAP with guideline-concordant antibiotic regimens, fluoroquinolone monotherapy is as effective as macrolide/beta-lactam combinations.<sup>15</sup> Importantly, non-adherence to CAP treatment guidelines is a significant risk factor for treatment failure and mortality.<sup>16</sup>

An assessment of existing national guidelines for the treatment of lower respiratory tract infections (LRTIs) and/or CAP in Europe was recently conducted by questionnaire sent to ERS national delegates.<sup>17</sup> The survey revealed that 18 of 24 responding delegates had national or regional guidelines for the management of CAP, and of those, seven guidelines included recommendations on the differential diagnosis, treatment, and management of TB. Seven responders also confirmed that their guidelines included recommendations on the use of fluoroquinolones in CAP and the risk of selecting fluoroquinolone-resistant *M. tuberculosis* in misdiagnosed patients. In several countries in Europe with low TB incidence, opportunities for physicians to investigate a TB patient are relatively rare and so there is a risk that TB is not considered as a potential diagnosis when a patient with an LRTI presents for consultation. Revision of national and regional guidelines for the management of LRTIs and/or CAP is therefore warranted, specifically to describe the need to consider the differential diagnosis of TB and highlight the potential risk of fostering fluoroquinolone resistance in TB patients who are misdiagnosed and do not receive appropriate therapy.

### 3. Diagnosis of CAP

Data from clinical studies illustrate that the differential diagnosis of TB from bacterial pneumonia is not straightforward.

In Asian countries, 1–7% of cases presenting as CAP were re-diagnosed as pulmonary TB.<sup>18</sup> Most of these patients were over 65 years of age with various comorbidities.<sup>18</sup> In contrast, studies in Africa have identified *M. tuberculosis* as the cause of pneumonia in approximately 30% of HIV-infected patients,<sup>19,20</sup> indicating a shift in the aetiology of pneumonia in severely ill patients immunocompromised with advanced HIV.

In the absence of a diagnostic 'gold standard', the diagnosis of CAP is based on demonstration of a new infiltrate on chest radiograph or other imaging technique in the presence of recently acquired respiratory signs and symptoms. Chest radiography of patients with cough and fever lasting 2–3 days due to bacterial pneumonia reveals an airspace infiltrate, in clear contrast to the cavitating lung lesions seen in patients with a history of cough for 3 months or longer accompanied by weight loss, which are typical of TB. Clinical findings do not, however, reliably predict radiologically confirmed pneumonia,<sup>21</sup> as features of TB may sometimes be quite similar to those of CAP among patients who experience symptoms at the early stage. In addition, the etiology cannot be simply differentiated clinically or radiologically and is undefined in approximately 50% of patients.

The presence of HIV influences the presentation of pulmonary infections and so complicates the diagnosis of CAP and TB, particularly in areas of high TB prevalence. In HIV-positive patients, lung characteristics identified by chest X-ray or computed tomography imaging together with clinical course (acute vs. chronic onset) can be helpful in suggesting the etiology. This has enabled an algorithm approach to the evaluation of hospitalized HIV-seropositive patients with suspected CAP to be recommended.<sup>22</sup>

### 4. Fluoroquinolone treatment for TB

Fluoroquinolones have considerable potential to treat TB due to their favourable pharmacokinetics and activity against the target pathogen. Later-generation fluoroquinolones including gatifloxacin, levofloxacin (750 mg/day), moxifloxacin (maximum 400 mg/day), and even ofloxacin, are suggested by the World Health Organization (WHO) as second-line anti-TB agents.<sup>23</sup> However, none is licensed for use in the treatment of drug-susceptible TB, and these should only be used for the treatment of multidrug-resistant TB (MDR-TB),<sup>24</sup> or when toxicity curtails the use of standard anti-TB therapy. Earlier fluoroquinolones (sparfloxacin and ciprofloxacin) have also been evaluated in some clinical trials, but are not generally considered effective as second-line agents. Ciprofloxacin should not be used.<sup>25</sup> While some small studies have indicated the efficacy of fluoroquinolones in TB,<sup>3–6</sup> no large-scale controlled clinical trial has been completed. In addition, these drugs are intended and approved for short-term use and safety data are lacking for their long-term use.

Moxifloxacin 400 mg is currently being tested in two phase III multicentre international clinical trials: the Rapid Evaluation of Moxifloxacin in the treatment of sputum smear positive Tuberculosis (REMoxTB) study and the International Multicentre Controlled Clinical Trial to Evaluate High Dose Rifapentine and a Quinolone in the Treatment of Pulmonary Tuberculosis (RIFAQUIN). Both studies are investigating the possibility of shortening chemotherapy from 6 to 4 months, which is expected to substantially improve treatment completion rates and adherence. In the REMoxTB study, one group is given 6 months of standard treatment, a second group receives moxifloxacin substituted for ethambutol as part of a 4-month regimen, and a third group receives moxifloxacin substituted for isoniazid as part of a 4-month regimen.<sup>26,27</sup> In the now completed RIFAQUIN study,<sup>28</sup> three drug combination regimens were compared. The 6-month control standard regimen contained rifampin, isoniazid, ethambutol, and

pyrazinamide, while the test regimen was given for 6 months or 4 months and contained rifampin, moxifloxacin, rifapentine, ethambutol, and pyrazinamide given daily in a 2-month intensive phase. The study results will demonstrate whether the new regimens (containing moxifloxacin) for 6 months or 4 months were non-inferior to standard therapy. A 4-month regimen containing gatifloxacin is also being tested in a different phase II clinical trial in five African countries. The test treatment comprises the standard combination of drugs with gatifloxacin in place of ethambutol administered daily for 2 months. During the continuation phase, patients will receive weekly treatment with gatifloxacin, rifampin, and isoniazid for 2 months.<sup>29</sup>

## 5. Use of fluoroquinolones in CAP in TB-endemic areas: current issues

Empiric treatment of CAP in areas with high TB prevalence has raised some questions regarding the use of fluoroquinolones:

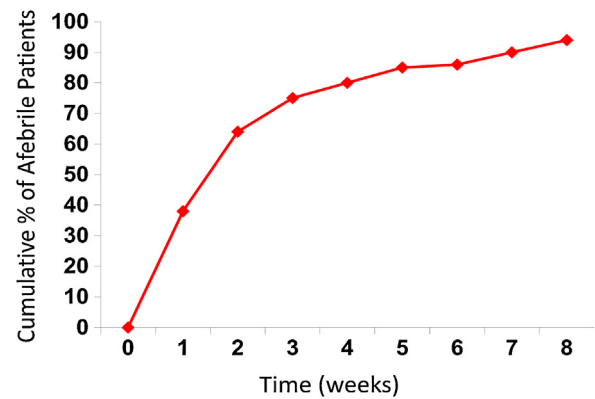
- (1) Among patients with an LRTI, does fluoroquinolone treatment delay the diagnosis of pulmonary TB?
- (2) If a patient with subsequently diagnosed TB improves rapidly with fluoroquinolone treatment, is the improvement truly an improvement of TB or resolution of a concurrent bacterial respiratory tract infection?
- (3) Is the eventual diagnosis of TB delayed?
- (4) If a delay in diagnosis occurs, does this affect outcome and is it specifically related to the use of fluoroquinolones?
- (5) Is the use of fluoroquinolones associated with a higher frequency of culture-negative TB?
- (6) If fluoroquinolones are used to treat LRTIs, does this induce fluoroquinolone resistance in *M. tuberculosis* isolates?
- (7) If so, what is the scale of exposure required?
- (8) What is the impact of the use of respiratory fluoroquinolones in the treatment of TB or CAP on the development of resistance in organisms other than *M. tuberculosis* (e.g., *S. pneumoniae*, *Enterobacteriaceae*)?

## 6. Clinical differentiation of CAP from TB

Comparing the typical clinical courses of CAP and TB provides some useful points of difference. For example, in a prospective observational study of time-to-clinical stability in patients hospitalized with CAP, fever was resolved (highest temperature for the day  $\leq 37.8^\circ\text{C}$ ) in a median of 3 days (interquartile (IQR) range 2–4 days).<sup>30–32</sup> In comparison, a study of patients with pulmonary TB who received appropriate multidrug anti-TB therapy found that fever resolved after a mean of 16 days (Figure 1). An important caveat to this observation is that antimicrobial treatment of a presumed co-existing bacterial infection did not influence the course of fever.<sup>33</sup>

In two-thirds of patients with bacterial pneumonia, radiological evidence of pulmonary infiltrates is absent 4 weeks after diagnosis.<sup>34</sup> Although radiographic monitoring of the response during TB treatment is not recommended,<sup>35</sup> radiography can be expected to show positive changes within 1 month and resolving or becoming stable in 90% of patients by 6 months.<sup>36</sup> Conversely, a small proportion of patients with TB can have progressive pulmonary infiltrates despite evidence of clinical improvement in response to appropriate antibiotic therapy.<sup>37</sup>

Clinical features predictive of pulmonary TB were identified in a prospective study in which *M. tuberculosis* was isolated from 4.9% of patients hospitalized for CAP. The presence of symptoms lasting more than 2 weeks prior to admission, upper lobe involvement or cavitary infiltrates on chest radiograph, total white blood cell count  $\leq 12 \times 10^9/\text{l}$  on admission, night sweats, and lymphopenia were all



**Figure 1.** Cumulative percentage of patients becoming afebrile during treatment for tuberculosis. (Reprinted with permission of the American Thoracic Society. Copyright © 2013 American Thoracic Society. Kiblawi SS, et al. Fever response of patients on therapy for pulmonary tuberculosis. *Am Rev Respir Dis* 1981;123:20–4. Official journal of the American Thoracic Society.).

significantly associated with culture-positive pulmonary TB.<sup>38</sup> Identification of these features at presentation clearly strengthens the diagnostic suspicion of TB, and sputa should be submitted for smear and culture analysis.

Taken together, these findings indicate that if a patient with symptoms of pneumonia responds quickly to antimicrobial therapy, they are likely to have a bacterial pneumonia. In contrast, TB is not associated with a rapid response to treatment even when treated with appropriate multidrug regimens.

### 6.1. Laboratory tests to differentiate CAP from TB

The tuberculin skin test (TST) has been the standard immuno-diagnostic test for TB for over a century and is still widely used in screening to detect the immune response to mycobacterial antigens, but is not useful as a method to diagnose the disease. The overall sensitivity of the TST has been estimated as 77% in a meta-analysis,<sup>39</sup> but the sensitivity can be substantially impaired by a variety of factors. In particular, the specificity of the TST is dependent on the *Bacillus Calmette–Guérin* vaccination status<sup>40</sup> and the immune status of the person being tested.<sup>39</sup> In the context of differentiating CAP and TB, the TST lacks the required sensitivity and specificity and is not recommended.<sup>41</sup>

Interferon gamma (IFN- $\gamma$ ) release assays (IGRAs), which measure T-cell release of IFN- $\gamma$  in response to *M. tuberculosis*-specific antigens, have high sensitivity for active TB, superior to that of the TST. However, the specificity of IGRAs is poor in patients with suspected active TB in high TB burden settings, suggesting they are of limited use as a confirmatory test for active TB in TB-endemic countries with a high background prevalence of latent TB.<sup>42</sup> A WHO Expert Group has discouraged the use of IGRAs for the diagnosis of active pulmonary TB in low- and middle-income countries.<sup>43</sup>

In resource-limited settings, TB diagnosis typically relies on the identification of acid-fast bacilli (AFB) on unprocessed sputum smears using conventional light microscopy. This approach has proved highly specific for pulmonary TB due to *M. tuberculosis* in high TB incidence areas. The overall sensitivity of sputum-based diagnosis is 20–80%,<sup>44</sup> and is highest for patients with cavitary disease and lowest in patients with weak cough or less advanced disease.<sup>44</sup> Diagnosis requires a concentration of bacilli of 5000–10 000/ml for a trained and skilled technician to detect 1–3 organisms in 300 oil immersion fields. The overall yield for smear and culture is superior with multiple specimens. Compared with conventional light microscopy, fluorescence microscopy is more sensitive and has similar

specificity.<sup>45</sup> Although a single sputum specimen is sufficient to establish the diagnosis by culture in HIV-positive patients, a minimum of two smears is needed to achieve an acceptable early diagnostic yield.<sup>46</sup> Repeated sputum induction considerably improves diagnostic accuracy.<sup>47</sup> Bronchoscopy is useful for patients with radiographic features consistent with TB but who have smear-negative sputum or produce no sputum.<sup>48</sup>

Nucleic acid amplification assays should be used to confirm the presence of *M. tuberculosis* following a smear test positive for AFB. The Xpert MTB/RIF assay (Cepheid Inc., Sunnyvale, CA, USA) can accurately detect TB and rifampin resistance in less than 2 h. The WHO-endorsed assay is a fully integrated and automated system that is simple to perform and requires minimal training and laboratory facilities. Studies have shown it to be sensitive and specific,<sup>49</sup> superior to AFB smear microscopy,<sup>50,51</sup> with high sensitivity in smear-negative TB<sup>51</sup> and effective for the early and accurate diagnosis of TB and MDR-TB in low-resource, TB-endemic settings.<sup>52</sup>

The low levels of serum C-reactive protein (CRP) and procalcitonin (PCT) found in patients with pulmonary TB provide useful discrimination between those with bacterial CAP, including HIV-positive patients, and those with pneumonia caused by *Pneumocystis jirovecii* infection.<sup>53–56</sup> A recent study in Korea, an intermediate TB-burden country, found the neutrophil-lymphocyte count ratio (NLR) to be significantly lower in patients with pulmonary TB than in those with bacterial CAP, and to provide superior diagnostic discrimination of the two diagnoses than CRP.<sup>57</sup>

The detection of *M. tuberculosis* antigens in urine represents an important potential approach for the diagnosis of TB in resource-limited settings. However, this method is not currently accepted as a gold standard in many low income countries. The lipoarabinomannan urinary assay shows most promise, but has suboptimal sensitivity for routine clinical use.<sup>58</sup> However, positive urinary antigen tests for pneumococcal and *Legionella* antigens allow early exclusion of TB.

## 7. Does fluoroquinolone treatment extend the delay to diagnosis that commonly occurs in cases of pulmonary TB?

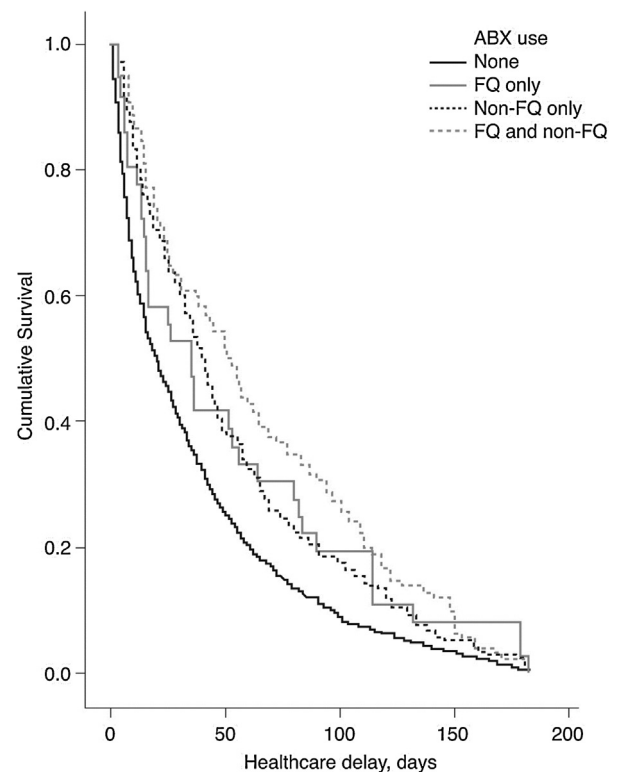
Delay in TB diagnosis can either be patient delay or healthcare system delay. Patient delay refers to the time from onset of clinical symptoms to the first visit to a healthcare centre, while healthcare system delay is the time from first patient visit to a healthcare centre to establishment of a TB diagnosis.<sup>59</sup> An average patient delay of 4 weeks and an average healthcare system delay of 3–5 weeks are common.<sup>59</sup> Even in hospitalized patients with smear-positive disease, delays in the suspicion and treatment of TB are common, with one study finding overall management delays of more than 10 days occurring in a third of patients.<sup>60</sup>

Several studies have investigated the influence of empirical antibiotic treatment for respiratory infection on the period from presentation to diagnosis of TB. A small retrospective cohort study in Baltimore, USA, showed a longer median time (16 days,  $p = 0.04$ ) between presentation and treatment of pulmonary TB in patients prescribed fluoroquinolones compared with those who received non-fluoroquinolone antibiotics.<sup>61</sup> A similar result was reported from a randomized open-label study in Hong Kong, although paradoxically the study data showed that of those patients who developed active pulmonary TB during a 1-year follow-up, 4.8% were given amoxicillin-clavulanate and 1.4% were given moxifloxacin.<sup>62</sup> A retrospective study in Taiwan identified longer duration from initial visit and from mycobacterial culture sampling to the start of anti-TB treatment in patients with confirmed TB who had received empirical therapy with fluoroquinolones. However, the patients' age, higher prevalence of

constitutional symptoms and malnourishment, and lower frequency of AFB-positive sputum tests, suggest that the different clinical presentation of these patients probably contributed to the different course of diagnosis and treatment.<sup>63</sup>

A meta-analysis of four studies<sup>61,63–65</sup> showed a mean duration of delayed diagnosis and treatment of pulmonary TB of 19 days in patients prescribed fluoroquinolones compared with those who received non-fluoroquinolone antibiotics. This analysis also showed, however, that the initiation of anti-TB antibiotics was not delayed in patients prescribed fluoroquinolones. Although intended to investigate the effect of prior antibiotic treatment for CAP, patients in the studies included those who had received fluoroquinolones for a variety of non-respiratory infections, including urinary tract infections and wound infections.<sup>66</sup>

The findings of a single, large, population-based study in British Columbia provide clearer evidence that healthcare delays with pulmonary TB patients occur following treatment with any antibiotic, not just with a fluoroquinolone<sup>67</sup> (Figure 2). Using the linked health databases of the province, this study collected data for 2232 patients who had active TB between 1997 and 2006. After excluding incomplete patient records, data were analyzed for 1544 patients with antibiotic exposure within 6 months prior to the initiation of anti-TB treatment, 414 of whom (27%) received antibiotics, while the remaining 1130 (73%) did not. Antibiotic-treated patients experienced on average more than twice the healthcare delay compared with the non-antibiotic group, after adjusting for covariates; the median healthcare delay was 41 days (IQR 15–86) for the antibiotic group compared with 14 days (IQR 3–44) for the non-antibiotic group (adjusted risk ratio (RR) 2.12, 95% confidence interval (CI) 1.82–2.46). When stratified by type of antibiotic use, there was no difference in the delay in diagnosis of TB between those who received non-fluoroquinolone antibiotics



**Figure 2.** Time to tuberculosis treatment from initial contact with healthcare services by antibiotic type. (Reprinted with permission of the International Union Against Tuberculosis and Lung Disease. Copyright © The Union. Wang M, et al. Is the delay in diagnosis of pulmonary tuberculosis related to exposure to fluoroquinolones or any antibiotic? *Int J Tuberc Lung Dis* 2011;15:1062–8.).



(adjusted RR 2.00, 95% CI 1.67–2.38), fluoroquinolones only (adjusted RR 2.18, 95% CI 1.42–3.32), and mixed fluoroquinolone and non-fluoroquinolone (adjusted RR 2.37, 95% CI 1.86–3.03) (Figure 2). Increased treatment delays were also related to the number of courses of antibiotics prescribed. These data suggest that the delay in initiating anti-TB treatment is more probably a result of diagnostic doubt. Consistent with this, Golub et al.<sup>64</sup> also found that diagnostic delays were associated with all classes of antibiotics prescribed and noted that when a physician considered the possibility of TB (e.g., requesting a sputum smear and mycobacterial culture, or receiving a radiograph report suggesting TB), antibiotics were less likely to be prescribed. Similarly, a UK study that found longer times to diagnosis of TB with prior antibiotic treatment revealed that the delay appeared to be a consequence of prolongation of the healthcare process and was not predicted by symptomatic improvement.<sup>68</sup> This suggests that a delayed diagnosis of TB may not be due to the anti-TB activity of fluoroquinolones, but rather the time inherent in taking a course of antibiotics and waiting to see if there is a clinical response. In contrast, Jeon et al.<sup>69</sup> reported that TB patients exposed to a fluoroquinolone for 5 days or more before sputum collection were more likely to be smear-negative than unexposed patients, and that this was likely to be mediated by the antibacterial effect of fluoroquinolones. However, fluoroquinolone use is not associated with an increased risk of culture-negative TB.<sup>70</sup>

## 8. Development of fluoroquinolone resistance in *M. tuberculosis*

The cellular target of fluoroquinolones in *M. tuberculosis* is DNA gyrase, a tetrameric type II topoisomerase composed of two A and two B subunits encoded by the *gyrA* gene and *gyrB* gene, respectively.<sup>71</sup>

Unlike many bacterial species, *M. tuberculosis* appears to lack topoisomerase IV, a cellular protein also inhibited by fluoroquinolones, and DNA gyrase appears to be the sole target for fluoroquinolone antibiotics.<sup>72</sup> Genetic resistance to an anti-TB drug is caused by spontaneous chromosomal mutations at a frequency of  $10^{-6}$  to  $10^{-8}$  mycobacterial replications. Mobile genetic elements such as plasmids and transposons, known to mediate drug resistance in various bacterial species, do not cause mutations in *M. tuberculosis*.<sup>73</sup>

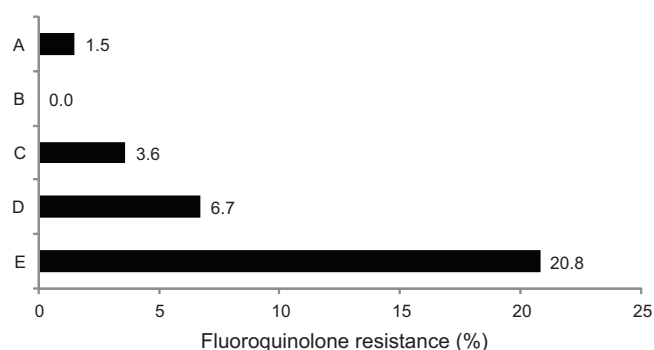
Fluoroquinolone resistance in *M. tuberculosis* is mainly due to the acquisition of point mutations within a conserved region of *gyrA* (320 bp) and *gyrB* (375 bp), the quinolone resistance-determining region (QRDR). Mutations within the QRDR of *gyrA* account for 42–100% of fluoroquinolone resistance in *M. tuberculosis*, with codons 90, 91, and 94 being the most mutated sites.<sup>73</sup> Resistance due to *gyrB* mutations was thought to be rare, but clinical isolates resistant to fluoroquinolones with *gyrB* mutations and wild-type *gyrA* loci have recently been reported in several studies.<sup>74</sup> In addition, the *M. tuberculosis* pentapeptide repeat protein MfpA mediates fluoroquinolone resistance by interacting with DNA gyrase and protecting it from antibiotic binding.<sup>75</sup> The contribution of MfpA expression and other mechanisms potentially responsible for clinical resistance of *M. tuberculosis* to fluoroquinolones, such as decreased cell wall permeability, drug efflux pump, drug sequestration, or drug inactivation, requires clarification.<sup>73</sup>

The emergence of MDR and XDR strains of *M. tuberculosis* reflects multiple aspects of inadequate TB management, including poor supervision of anti-TB treatment; the misuse of isoniazid and rifampin, for example, has been widespread.<sup>76</sup> The effectiveness of standard TB therapy can be compromised by several factors, including poor adherence associated with adverse events and long duration of treatment, or inadequate drug levels for other reasons

(e.g., drug–drug interactions, poor quality medicines, use of over-the-counter antibiotics). These factors increase the risk of unsuccessful treatment outcomes and the development of drug resistance to one or more of the drugs in the regimen.<sup>77,78</sup> With isoniazid monotherapy, for example, the emergence of resistance in *M. tuberculosis* is uncommon during the first 3 months of treatment, but more frequent with continuing monotherapy.<sup>79</sup>

An early study in New York City identified 22 patients with fluoroquinolone-resistant *M. tuberculosis*, 16 of whom had received ciprofloxacin or ofloxacin. The median (range) time between isolation of a fluoroquinolone-susceptible strain and a fluoroquinolone-resistant strain was 137 (43–398) days after a period of fluoroquinolone treatment of 64 (23–271) days,<sup>80</sup> far longer than the recommended treatment course for CAP. Fluoroquinolone resistance in two of 55 patients with TB (4%) was reported by a small US study. Both patients had had fluoroquinolone treatment within the previous 3 months; both were also HIV-seropositive with low CD4+ lymphocyte counts, reflecting poor immunity.<sup>71</sup>

Important insights into the prevalence of and risk factors for fluoroquinolone resistance in *M. tuberculosis* were reported by a study in Tennessee, USA.<sup>81</sup> Of 1136 culture-confirmed cases, 640 had isolates available for fluoroquinolone susceptibility testing; those with fluoroquinolone-resistant isolates were compared with those with susceptible isolates. Of the 640 study patients, 116 (18%) had received fluoroquinolones as outpatients before the diagnosis of TB and 54 (8.4%) had received fluoroquinolones for more than 10 days. Sixteen patients (2.5%) had fluoroquinolone-resistant *M. tuberculosis* isolates. Regression analyses revealed that >10 days fluoroquinolone exposure was associated with fluoroquinolone-resistant TB, while age, gender, race, and HIV serostatus were not associated with fluoroquinolone resistance. In addition, patients receiving more than one course of fluoroquinolone treatment were more likely to have fluoroquinolone-resistant TB than those who received only one course ( $p = 0.007$ ). Assessment of the duration and timing of the last fluoroquinolone exposure showed that patients receiving fluoroquinolone therapy of  $\geq 10$  days duration more than 60 days before the diagnosis of TB had the highest proportion (20.8%) of fluoroquinolone-resistant TB (Figure 3).



**Figure 3.** Percent fluoroquinolone resistance according to duration of fluoroquinolone exposure ( $\leq 10$  days vs.  $>10$  days) and timing of last exposure ( $\leq 60$  days vs.  $>60$  days) before tuberculosis diagnosis. (A) No outpatient fluoroquinolone exposure ( $n = 524$ ). (B)  $\leq 10$  days of fluoroquinolones and last fluoroquinolone exposure  $\leq 60$  days before tuberculosis diagnosis ( $n = 34$ ). (C)  $\leq 10$  days of fluoroquinolones and last fluoroquinolone exposure  $>60$  days before tuberculosis diagnosis ( $n = 28$ ). (D)  $>10$  days of fluoroquinolones and last fluoroquinolone exposure  $\leq 60$  days before tuberculosis diagnosis ( $n = 30$ ). (E)  $>10$  days of fluoroquinolones and last fluoroquinolone exposure  $>60$  days before tuberculosis diagnosis ( $n = 24$ ). (Reprinted with permission of the American Thoracic Society. Copyright © 2013 American Thoracic Society. Devasia RA, et al. Fluoroquinolone resistance in *Mycobacterium tuberculosis*: the effect of duration and timing of fluoroquinolone exposure. Am J Respir Crit Care Med 2009;180:365–70. Official journal of the American Thoracic Society.).

In a case–control study in Canada, Long et al.<sup>82</sup> found that multiple but not single prescriptions of fluoroquinolones were associated with fluoroquinolone-resistant TB. This association was also true for *M. tuberculosis* strains resistant to first-line anti-TB treatment. Three strains of *M. tuberculosis* isolated from cases had increased MICs for ciprofloxacin, levofloxacin, and ofloxacin, although only one strain had a resistance-conferring *gyrA* mutation. These three strains were isolated from patients who had received multiple ciprofloxacin treatments.<sup>82</sup> Similarly, a retrospective study in South Africa found that one of 201 genotyped *M. tuberculosis* isolates harboured a resistance-conferring *gyrA* mutation. This isolate was obtained from a patient who had been exposed to a total of 8 days of fluoroquinolone treatment given over three different intervals before culture collection: 1 day of ciprofloxacin 79 days prior, 2 days of ofloxacin 42 days prior, and 5 days of ciprofloxacin 5 days prior.<sup>69</sup>

Park et al.<sup>83</sup> reported the frequencies of ofloxacin resistance as 1.1% in patients with no recent exposure to fluoroquinolones and 8.5% in those who received fluoroquinolone monotherapy within the previous 3 months. Ofloxacin resistance usually accompanied multidrug resistance. In this study of 2788 Korean patients from 1997 to 2005, the median (range) duration of fluoroquinolone treatment was 7 days (1–47 days) and 35 of 39 patients received at least 5 days of fluoroquinolone therapy before *M. tuberculosis* culture was performed.<sup>83</sup> In contrast, a study in Taiwan found that neither the previous use of fluoroquinolones nor the duration of fluoroquinolone exposure was correlated with the fluoroquinolone susceptibility of *M. tuberculosis* isolates, 3.3% of which were fluoroquinolone-resistant.<sup>84</sup> However, this study of patients in tertiary care did not have access to data on previous medication history, including prior fluoroquinolone use, outside the hospital. Resistance to fluoroquinolones was also correlated with prior anti-TB treatment and with resistance to any first-line anti-TB drug (isoniazid, rifampin, and ethambutol).

The association between higher rates of fluoroquinolone resistance amongst MDR *M. tuberculosis* strains compared with susceptible strains is supported by a recent analysis of the frequency of and risk factors for acquired resistance to second-line drugs using data from the US National Tuberculosis Surveillance System 1993–2008. This analysis identified MDR-TB at treatment initiation as the only predictor for acquired resistance to fluoroquinolones (ofloxacin or ciprofloxacin).<sup>85</sup>

The clinical data reviewed above strongly suggest that fluoroquinolone resistance in TB requires repeated and/or prolonged courses of monotherapy and is associated with the presence of MDR rather than previous fluoroquinolone exposure.

In addition to the delay in TB diagnosis and risk of fluoroquinolone resistance, the risk of mortality may also be influenced by a previous course of fluoroquinolone treatment for patients in areas where TB is not highly endemic.<sup>86</sup> A study by van der Heijden et al. showed an increased risk of mortality (OR 1.82) if patients were exposed to fluoroquinolones (ciprofloxacin, levofloxacin, or moxifloxacin) before a correct diagnosis of TB. However, the association between fluoroquinolone exposure and mortality was not present without adjusting for comorbidities such as chronic obstructive pulmonary disease, diabetes mellitus, or alcoholism, and when patients with unknown HIV status were excluded from the analysis.<sup>86</sup>

## 9. Conclusions

The use of fluoroquinolones for 5–10 days as empirical treatment for CAP, according to current clinical management guidelines, is appropriate even in regions endemic for TB. However, indiscriminate use of fluoroquinolones in suspected CAP should be avoided, and critical judgement on the possibility of TB must take

place when patients with LRTIs visit respiratory physicians. TB should always be considered by the physician as a possible cause of pneumonia, however, and if suspected, the relevant diagnostic tests should be completed rapidly before prescribing CAP-directed antibiotics. Empiric antibiotic treatment for suspected CAP (e.g., fluoroquinolone monotherapy) should not be started for patients with a protracted LRTI associated with cough, fever, and weight loss together with cavitary lung lesions without first excluding TB. Earlier-generation fluoroquinolones such as ciprofloxacin and ofloxacin should be avoided for both CAP and TB, as they have lower activity against both *S. pneumoniae* and *M. tuberculosis*. Similarly, if a patient with symptoms of pneumonia does not respond to a short course of empiric antibiotic therapy, that therapy should not be continued and further pathologies, including possible TB, should be investigated; prolonged and/or repeated courses of fluoroquinolone monotherapy may be associated with the emergence of fluoroquinolone resistance in *M. tuberculosis* and/or increased mortality. In patients with LRTIs who subsequently develop TB, empiric therapy with any antibiotic can delay the diagnosis of TB. However, delays in the diagnosis of TB are common, even without empiric antibiotic treatment, including fluoroquinolones. This reflects the diagnostic doubt often inherent in such cases. Fluoroquinolones are important drugs for the treatment of MDR-TB but should not be used in susceptible disease until the results of ongoing clinical trials are available or in the case of drug toxicity.

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